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A New Species of *Chirixalus* from Vietnam (Anura: Rhacophoridae)

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ABSTRACT—A new rhacophorid species is described on the basis of two specimens collected from Vu Quang Nature Reserve, Ha Tinh Province, central Vietnam. The species has inner and outer fingers that are not opposable, but in order to avoid taxonomic confusion, it is tentatively assigned to the genus *Chirixalus*. It is a large *Chirixalus*, having robust body with warty, grayish dorsum and immaculate ventrum, and lacking large pollex, white granules around anus and on limbs, and dark markings on sides of body. It is most similar to *C. eiffingeri* and *C. idiootocus* in external morphology, and much different from the other congeners. Generic definition of the genera *Chirixalus* and *Kurixalus* is discussed.

Key words: classification, taxonomy, systematics, biogeography, Southeast Asia

INTRODUCTION

Liem (1970) made an extensive morphological study of the Old World tree frogs and established a taxonomy to split members of the family Rhacophoridae into ten genera. Since then several new suprageneric classifications have been proposed by subsequent authors (e.g., Channing, 1989; Dubois, 1981, 1992; Bossuyt and Dubois, 2001), and many new species have been added to some genera (e.g., *Rhacophorus*, *Polypedates*, and *Philautus*: Inger *et al.*, 1999; Manamendra-Arachchi and Pethiyagoda, 2001; Orlov *et al.*, 2001; Ziegler und Köhler, 2001). The genus *Chirixalus* Boulenger, 1893, however, is limited in the range of distribution and small in the number of species included, as shown by the fact that few new members have been added to the genus (Glaw *et al.*, 1998).

In Vietnam, extensive field surveys by many groups of herpetologists have resulted in the discovery of new frog taxa of different lineages including rhacophorid genera *Philautus* and *Rhacophorus* (Inger *et al.*, 1999; Ohler *et al.*, 2000; Orlov *et al.*, 2001; Ziegler und Köhler, 2001). Many undescribed species, however, are actually still present in this country, and during our survey of the central region in 1997, we collected a pair of specimens of a rhacophorid distinctly different from the species hitherto known to occur in

Vietnam (Bourret, 1942; Inger *et al.*, 1999). Although allocations of a species into a known genus of rhacophorids include severe taxonomic problems (see Discussion), we assign these specimens to the genus *Chirixalus* and describe them below as a new species.

MATERIALS AND METHODS

The junior author conducted field work at Vu Quang Nature Reserve, Rao Cua District, Ha Tinh Province, central Vietnam in June of 1997. Specimens were fixed in 10% formalin and later preserved in 70% ethanol. For preserved specimens, we took 18 body measurements (Table 1), mainly following Matsui (1984), to the nearest 0.1 mm with dial calipers under a binocular dissecting microscope. Webbing formula is that of Myers and Duellman (1982). We prepared radiographs to examine gross osteology, and made slight dissections to examine gonads. For comparisons, we examined museum specimens, including types, of most described taxa of the genus *Chirixalus* and allied genera (Appendix 1).

SYSTEMATICS

Chirixalus ananjevae sp. nov.
(Figs. 1, 2)

Holotype

KUHE (Kyoto University, Human and Environmental Studies) 31963, an adult male from Vu Quang Nature Reserve, Rao Cua District, Ha Tinh Province, Vietnam (18°16'N, 105°28'E, alt. 1500 m), collected on 16 June 1997 by Nikolai Orlov.

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Table 1. Measurements in mm. SVL: snout-vent length; HL: head length (= from tip of snout to hind border of the angle of jaw, measured not parallel with the median line); SL: snout length; EL: eye length; T-EL: tympanum-eye length; TD: tympanum diameter; HW: head width; IND: internarial distance; IOD: interorbital distance; UEW: upper eyelid width; FLL: forelimb length; 3FDW: third finger disk diameter; HLL: hindlimb length; TL: tibia length; FL: foot length (= from proximal end of inner metatarsal tubercle to tip of fourth toe); 4HDW: fourth toe disk diameter; 1TL: first toe length (= from distal end of inner metatarsal tubercle to tip of first toe); IMTL: inner metatarsal tubercle length.

	Male	(%SVL)	Female	(%SVL)
SVL	32.4	—	43.4	—
HL	11.6	(35.8)	14.7	(33.9)
SL	4.9	(15.1)	5.7	(13.1)
EL	4.9	(15.1)	5.8	(13.4)
T-EL	0.7	(2.2)	1.1	(2.5)
TD	2.1	(6.5)	2.6	(6.0)
HW	12.1	(37.3)	15.7	(36.2)
IND	3.4	(10.5)	4.4	(10.1)
IOD	3.8	(11.7)	4.2	(9.7)
UEW	3.1	(9.6)	3.7	(8.5)
FLL	21.7	(67.0)	27.4	(63.1)
3FDW	1.6	(4.9)	2.5	(5.8)
HLL	49.3	(152.2)	62.0	(142.9)
TL	15.6	(48.1)	19.9	(45.9)
FL	14.3	(44.1)	18.8	(43.3)
4HDW	1.5	(4.6)	1.9	(4.4)
1TL	3.8	(11.7)	5.5	(12.7)
IMTL	1.6	(4.9)	1.8	(4.1)

Paratype

ZISP (Zoological Institute, St. Petersburg) 6756, an adult female, data same as the holotype.

Diagnosis

A large *Chirixalus*, male 32 mm, female 43 mm in SVL; pupil horizontal; vomerine teeth absent; inner and outer fingers not opposable; two outer metatarsals separated with webbing; vocal pouch not loose or transparent, skin of throat plain, without gular gland; Wolffian duct along the kidney simple, lacking short branches; bottle-shaped vesicula seminalis absent; nuptial pad present in male; eggs bicolored, small in diameter, and large in number; vertebrae procoelus, contour of the centrum nearly cylindrical; vertebral column moderately elongated; frontoparietal only slightly trapezoidal, flared laterally, and lacking parieto-squamosal arch; distal end of third metacarpal dilated distally with a disto-medial bony knob; distal end of the terminal phalanx slightly bifurcate and not Y-shaped; second, third, and fourth tarsals fused; like *C. eiffingeri* or *C. idiootocus* in body shape and grayish dorsal color, but differing from *C. eiffingeri* in lacking vomerine teeth or large pollex, and from *C. idiootocus* in having less pointed snout and larger body, and in lacking white granules around anus and on limbs, and dark markings on sides of body; differing from *C. doriae*, *C. vittatus*, *C. hansenae*, *C. laevis*, and *C. nongkhorensis*, by more robust body with larger head and less elongate trunk, as well as by warty dorsum and bifurcated dark dorsal markings, and from *C. simus* and *C. dudhwaensis* by larger body, longer snout, and immaculate ventrum.

Description of holotype (measurements in mm)

Body robust (Fig. 1A, B), snout-vent length 32.4; Head (11.6) slightly shorter than wide (12.1); snout (4.9) as long as eye (4.9), very slightly pointed, projecting slightly; canthus blunt; lores oblique, slightly concave; nostril nearer to tip of snout than to eye; internarial distance (3.4) slightly narrower than interorbital (3.8); latter wider than eyelid (3.1); eye diameter much larger than eye-nostril (2.6); pupil horizontal; tympanum distinct, length (2.1) three-sevenths eye diameter and separated from eye by one-third of tympanum

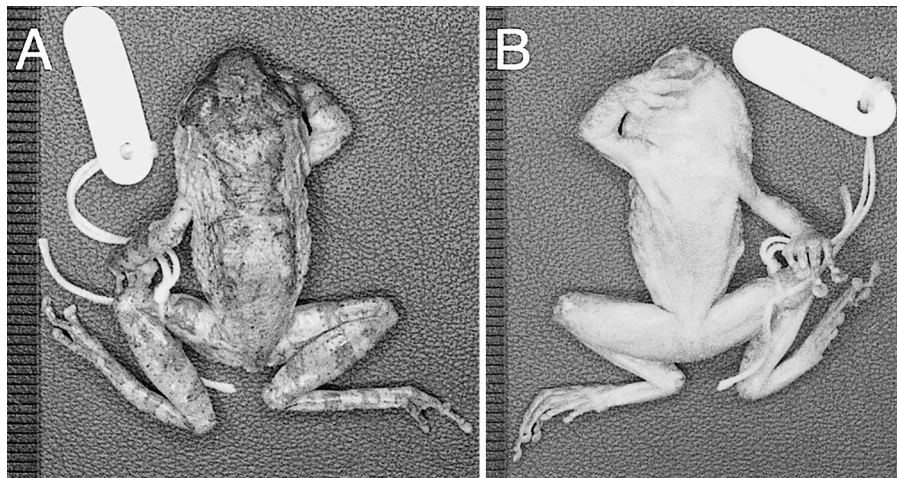


Fig. 1. Dorsal (A) and ventral (B) views of male holotype (KUHE 31963).

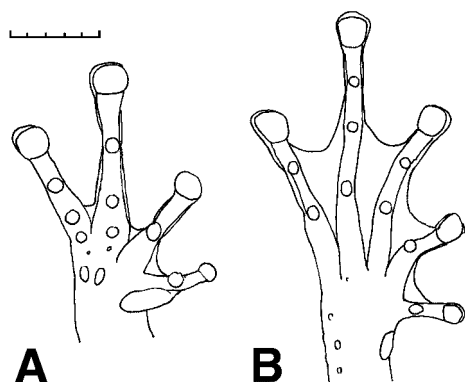


Fig. 2. Ventral views of hand (A) and foot (B) of male holotype (KUHE 31963). Scale bar indicates 5 mm.

diameter (0.7); vomerine teeth absent; a longitudinal opening into median subgular vocal sac on both sides of mouth floor.

Inner and outer fingers not opposable; first finger much shorter than second, length of first (2.5, measured from distal edge of inner palmar tubercle) about half diameter of eye; fourth finger much longer than second; tips of fingers dilated into large disks, that of third finger (1.6) narrower than tympanum; webs between fingers poorly developed (Fig. 2A), webbing formula $I2-2II2-basalllI3-2.75IV$; no fringe of skin on outer edge of fourth finger; a supernumerary tubercle on each metacarpal; subarticular tubercles distinct; distinct inner and two indistinct, elongate outer palmar tubercles.

Hindlimb (49.3) about 2.3 times length of forelimb (21.7); tibia not long (15.6), heels overlapping when limbs are held at right angles to body; tibiotarsal articulation of adpressed limb reaching middle of eye; foot (14.3) shorter than tibia; tips of toes expanded into disks slightly smaller than those of fingers (disk diameter of fourth toe 1.5); webs between toes moderately developed, two outer metatarsals separated with webbing (Fig. 2B); webbing formula $I1.5-2.7II1-2.7III1.5-3IV2.5-1V$; no fringes of skin along inner edge of first toe or outer edge of fifth; subarticular tubercles distinct; a small inner metatarsal tubercle, length (1.6) about two-fifths length of first toe (3.8), but no outer metatarsal tubercle.

Skin free of skull; dorsum with irregularly distributed small tubercles, those on snout small and white at tip; a distinct, oblique fold from eye, above tympanum, ending at shoulder; sides and abdomen coarsely granular; no ridge of skin on outer edge of forearm; leg smooth, except for coarsely granular ventral side of thigh and asperities on base of tarsus; no dermal appendages on limbs or at vent. Skin of throat plain, gular gland absent. Through the skin, tip of finger bones is not Y-shaped.

Cream-colored nuptial pad covering dorsomedial area on first finger from base to level of subarticular tubercle; Wolffian duct along the kidney a simple tube, lacking short branches and bottle-shaped vesicula seminalis.

Color in alcohol

Dorsum pinkish gray, with dark brown mark beginning between eyes, bifurcating at the back of shoulders, and ending on sides of sacrum (Fig. 1A); ventrally grayish white, without markings (Fig. 1B); limbs with dark crossbars; rear of thigh without distinct markings.

Osteology

Vertebrae procoelus; contour of centrum nearly cylindrical; relative length of vertebral column, expressed as the ratio of vertebral column length/greatest width of transverse process of the eighth vertebra, 2.2; frontoparietal only slightly trapezoidal, lacking parieto-squamosal arch; distal end of third metacarpal dilated distally with a disto-medial bony knob; distal end of terminal phalanx slightly bifurcate but not pointed, length of each branch less than width of phalanx; second, third, and fourth tarsals fused.

Variation

Snout-vent length of the female (43.4 mm) is larger than that of the male (32.4 mm). In the female paratype, tip of snout is rounded and internarial distance is slightly wider than interorbital. The hindlimb is proportionately shorter than the male and tibiotarsal articulation of adpressed limb reaches only to hind corner of eye. Unlike the male, the female lacks small asperities on snout or on base of tarsus. Instead, the female has warts on dorsal margin of vent. Otherwise the sexes are very similar in appearance.

Range

Known only from the type locality, Rao Cua District, Ha Tinh Province, Vietnam.

Natural history

In the middle of June, *C. ananjevae* was found in amplexus and perching on leaves of bush at night. No tadpoles or eggs were found in the pond and calling males were absent. The female contained many mature ova in the ovaries. The diameter of five ova ranged from 1.3–1.5 (mean \pm 2SE=1.39 \pm 0.05) mm. The animal pole is grayish brown and the vegetal pole is cream in color.

Etymology

The specific name is dedicated to Prof. Natalia Ananjeva of Russian Academy of Sciences who is always supporting our study.

DISCUSSION

Dubois (1981) reexamined Liem's (1970) results and split the genus *Philautus* Gistel, 1848, from all other members of Rhacophoridae and erected a monotypic subfamily Philautinae. He, however, later relegated the family Rhacophoridae to a subfamily of Ranidae and recognized three tribes (Dubois, 1992). By this action, Philautinae (Dubois, 1981) was changed to the tribe Philautini, which encom-

passes three subgenera. Regardless of this taxonomic change, the genus *Philautus* is defined by unique characters in internal and external morphology (see Liem, 1970 for details). One unique generic feature of external morphology is the absence or slight development of web between two outer metatarsals (Dubois, 1981; Manthey and Grossmann, 1997). The genus, however, is more clearly characterized by its unique reproductive habit of direct development from large yolk egg (Dubois, 1992, 1999). Unlike *Philautus*, the present new species has a web which extends between two outer metatarsals. Moreover the female possesses relatively large number of small eggs that are different from eggs of species with direct development.

The other six east Asian genera of Rhacophoridae (*Buergeria*, *Chirixalus*, *Nyctixalus*, *Polypedates*, *Rhacophorus*, and *Theloderma*), once grouped into the subfamily Rhacophorinae (Dubois, 1981), were divided into two tribes (Buergerini and Rhacophorini) including five genera (the genus *Polypedates* was synonymized with *Rhacophorus*: Dubois, 1992). The tribe Buergeriini (Channing, 1989) is monotypic, including the genus *Buergeria* which inhabits streams and has long hindlimbs with broad webs between toes (Maeda and Matsui, 1999). From the poor development of webs on toes and short hindlimb, the present new species is not considered to be a member of *Buergeria*, although we have no information of its breeding habits. Further, diplasio-coelous vertebral column and presence of vomerine teeth in *Buergeria* preclude the present new species from the genus.

Of the genera of the tribe Rhacophorini, the present new species is not assigned to *Nyctixalus*, because the pupil is not vertical. The new species is included neither in *Rhacophorus* (including *Polypedates*) nor in *Theloderma*, because the tip of finger bones is not Y-shaped (Manthey and Grossmann, 1997; Fei, 1999).

The remaining genus, *Chirixalus*, was proposed by Boulenger (1893:340) on the basis of *C. doriae* from Myanmar. Among diagnoses of the genus given by him, the lack of vomerine teeth and presence of fingers with the two inner opposed to the two outer seem to be unique. However, the former feature is currently considered to be variable within a genus of rhacophorids such as *Philautus* and *Rhacophorus* (Liem, 1970). The latter feature has been repeatedly listed as a diagnostic character of the genus, and Liem (1970) also noted it without actual character analyses. One important problem made by Liem (1970) is that he added in this genus *C. eiffingeri* (Boettger, 1895) from the Ryukyu Archipelago, Japan, and Taiwan, which actually lacks opposable fingers.

As if reflecting this discrepancy, *C. eiffingeri* has recently been separated from *Chirixalus* to represent a monotypic genus *Kurixalus* by Ye, Fei, and Dubois (Fei, 1999:383). This genus was defined by several morphological characteristics of adults, including weak development of vomerine teeth, non-opposable inner and outer fingers, and extremely developed pollex, but more important feature of *Kurixalus* is unique morphology and ecology of larvae that

are related to oviphagy.

Their classification (Ye, Fei, and Dubois in Fei, 1999), however, includes a problem, because a sister species of *C. eiffingeri*, *C. idiootocus* Kuramoto and Wang, 1987, from Taiwan, was not included in the genus *Kurixalus*, but was moved to *Philautus* without any reason. These two species are generally very close morphologically and differ from each other chiefly in the condition of pollex in adults, and in larval biology, with *C. idiootocus* having normal, non-enlarged pollex and normally feeding larvae (Kuramoto and Wang, 1987). They are also estimated to form a genetically unique lineage (Richards and Moore, 1998) and are suggested to be placed together in *Kurixalus*, (Wilkinson *et al.*, 2002).

The present new species from Vietnam is closer to *C. eiffingeri* and *C. idiootocus* than any other rhacophorids in external morphology and might also be included in *Kurixalus*. However, it is premature to recognize *Kurixalus* as a good genus distinct from *Chirixalus* because of two reasons. First, the genus *Kurixalus*, after inclusion of *C. idiootocus* and *C. ananjevae*, needs definition by clear synapomorphic characters, but this has never been done. Second, the genus *Chirixalus* itself is now considered to be polyphyletic (Wilkinson *et al.*, 2002), but no taxonomic assignment has been made as yet. Therefore, setting the problems of generic definition aside, it would at present be better to place the present new species in the genus *Chirixalus* (*sensu lato*) so as to avoid further taxonomic confusion until more extensive taxonomic revision of *Chirixalus* is made.

The present new species, *C. ananjevae* is similar to *C. eiffingeri* and *C. idiootocus* in the absence of opposable inner and outer fingers. However, it clearly differs from *C. eiffingeri* in the absence of vomerine teeth or development of pollex, and from *C. idiootocus* in less pointed snout, absence of vomerine teeth, and larger body. Additionally, *C. idiootocus* has white granules around anus and on limbs, and dark markings on sides of body, none of which are present in *C. ananjevae*. The remaining members of the genus (Glaw *et al.*, 1998) are reported to have opposable fingers, and are easily distinguished by external appearance from *C. ananjevae*. *Chirixalus doriae* Boulenger, 1893 and *C. vittatus* (Boulenger, 1887) from Myanmar to China, including Vietnam, *C. hansenae* (Cochran, 1927) from Thailand, *C. laevis* (Smith, 1924) from Vietnam, and *C. nongkhorensis* (Cochran, 1927) from Myanmar and Thailand, have more slender body with smaller head and more elongate trunk, as well as smoother back than *C. ananjevae*. The color and pattern of markings on dorsum are also much different. *Chirixalus simus* Annandale, 1915, from Assam, India, is similar to *C. ananjevae* in rugose dorsal skin and dorsal coloration, but is distinctly different from it in its smaller body size (22 mm SVL), shorter snout, longer hindlimb, and yellowish ventrum with clear markings. Another Indian species, *C. dudhwaensis* Ray, 1992, from Uttar Pradesh is very similar to *C. simus* (Ray, 1992; Das, personal communication). Neither do we consider here several

other species transferred from other genera to *Chirixalus* by Dubois and Ohler (2001), because their list (Dubois and Ohler, 2001: Table 6) includes some misidentifications (e.g., *Philautus romeri* Smith, 1953 from Hong Kong, which actually exhibits direct development and should remain placed in *Philautus*).

It is very interesting biogeographically, if Vietnamese *C. ananjevae* is actually phylogenetically close to two Taiwanese species, because no other amphibians exhibit such a distribution pattern. However, another rhacophorid genus, *Buergeria*, occurs from Japan to Taiwan and in Hainan Island (Maeda and Matsui, 1999), and this distribution may support the close relationships of Vietnamese and Taiwanese *Chirixalus*. In order to ascertain their relationships, future biochemical analyses would play a great role, although many problems have been only partially resolved so far by the molecular approach as shown by low supports of many clades shown by Wilkinson *et al.* (2002). Along with further biochemical information, data for natural history, especially for breeding habits, are also badly needed.

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Appendix 1 Specimens of rhacophorids examined for comparisons. For genera other than *Chirixalus*, only species whose types were examined are shown. Acronyms used are: BM=Natural History Museum, London; KUHE=Graduate School of Human and Environmental Studies, Kyoto University, Kyoto; MNHNP=Museum National d'Histoire Naturelle, Paris; MSNG=Museo Civico di Storia Naturale, Genova; NHMB=Naturhistorisches Museum Basel, Basel; NHMW=Naturhistorisches Museum Wien, Wien; OMNH=Osaka Museum of Natural History, Osaka; RMNH=Nationaal Natuurhistorisch Museum, Leiden; SMF=Forschungsinstitut und Naturmuseum Senckenberg, Frankfurt a. M.; ZMB=Universitat Humboldt, Zoologisches Museum, Berlin.

Chirixalus doriae Boulenger, 1893: NHMB 1247=possible paralectotype, KUHE 19025, 19065–66, 23713, 23720; *C. eiffingeri* (Boettger, 1895): SMF 6737=holotype, KUHE 10266–67, 12910; *C. hansenae* (Cochran, 1927): KUHE 34058, 34136–38; *C. idiootocus* Kuramoto and Wang, 1987: OMNH 9159–60, 10945–62, 84207–84226=paratypes; *C. nongkorensis* (Cochran, 1927): KUHE 19498–501, 19768, 19526–27; *C. vittatus* (Boulenger, 1887): KUHE 19178, 19193, 19208–09, 19272, 19441–48; *C. pollicaris* (Werner, 1914): NHMW 22881=type, 22882=cotype. *Buergeria buergeri* (Temminck and Schlegel, 1838): RMNH 1692=lectotype. *Philautus acutus* Dring, 1987: BM 1978.1765=holotype; *P. annandalii* (Boulenger, 1906): BM 1947.2.26.58=syntype; *P. aurifasciatus* (Schlegel, 1837): MNHNP 4590, 1989.3413=syntypes; *P. banaensis* Bourret, 1939: MNHNP 1948.160=lectotype, 1948.159, 161, 162=paralectotypes; *P. carinensis* (Boulenger, 1893): ZMB 11576, NHMB 1242–45=paratypes; *P. castanomerus* (Boulenger, 1912): BM 1947.2.26.88=holotype; *P. gryllus* Smith, 1924: BM 1947.2.5.95=holotype; *P. hosii* (Boulenger, 1895): BM 1947.2.8.89=holotype; *P. ingeri* Dring, 1987: BM 1978.1822–23=paratypes; *P. jerdonii* (Guenther, 1875): BM 1947.2.7.84–85=syntype; *P. larutensis*: BM 1947.2.6.36–38=syntype; *P. maosonensis* Bourret, 1937: MNHNP 1948.158=lectotype, 1948.157=paralectotype; *P. mjobergi* Smith, 1925: BM 1947.2.27.13=holotype; *P. palpebralis* Smith, 1924: BM 1947.2.4.46=holotype; *P. parvulus* (Boulenger, 1893): BM 1947.2.24.14–15=syntypes; *P. petersi* (Boulenger, 1900): BM 1947.2.27.16–17, 20=syntypes; *P. pulcherrimus* (Ahl, 1927): NHMB 1238=possible paratype. *Polypedates braueri* (Vogt, 1911): ZMB 21945=syntype; *P. teraiensis* (Dubois, 1987): MNHNP 1983.1103=holotype, 1983.1101, 1104, 1107, 1108, 1110, 1111, 1115, 1116, 1118, 1119=paratypes; *P. zed* (Dubois, 1987): MNHNP 1983.1141=holotype, 1983.1134–1140=paratypes. *Rhacophorus angulirostris* Ahl, 1927: MNHNP 1889.250–254=syntypes; *R. annamensis* Smith, 1924: BM 1947.2.8.86=holotype; *R. appendiculatus* (Guenther, 1859): 1947.2.9.17=syntype, BM 1947.2.8.99=holotype of *R. chaseni* Smith, 1924; *R. barbouri* Ahl, 1937: ZMB 11535=holotype; *R. calcaneus* Ahl, 1927: BM 1947.2.9.18=holotype; *R. cavirostris* (Gunther, 1868): BM 1947.2.31.29=holotype of *Ixalus fimbriatus* Gunther, 1872; *R. dorsoviridis* Bourret, 1937: MNHNP 1948.149=holotype; *R. edentulus* Mueller, 1894: BM 1947.2.8.82=paralectotype; *R. exiguus* Boettger, 1894: SMF 6987=holotype; *R. fasciatus* Boulenger, 1895: BM 1947.2.8.92–93=syntypes; *R. javanus* Boettger, 1893: SMF 6982=holotype; *R. macroscelis* Boulenger, 1896: BM 1947.2.8.85=holotype; *R. modestus* Boulenger, 1920: BM 1947.2.7.76–77=syntypes; *R. monticola* Boulenger, 1896: NHMB 1188=lectotype, NHMB 1190, 2351=paralectotypes; *R. notater* Smith, 1924: BM 1947.2.8.83=holotype; *R. phyllopygus* Werner, 1900: NHMB 1187=holotype; *R. poecilnotus* Boulenger, 1920: BM 1947.2.8.55–59=syntypes; *R. pulchellus* Werner, 1900: NHMB 1186=holotype; *R. reinwardtii lateralis* Werner, 1900: NHMB 1192=holotype; *R. taroensis* Smith, 1940: BM 1947.2.8.17=holotype; *R. turpes* Smith, 1940: BM 1947.2.8.69–70=syntypes; *R. verrucosus* Boulenger, 1893: MSNG 29853A=lectotype, MSNG 29853=paralectotype, BM 1944.2.8.91=paralectotype; *R. wirzi* (Roux, 1927): NHMB 3898=holotype. *Theloderma asperum* (Boulenger, 1886): BM 1947.2.6.18–19=syntypes; *T. bicolor* (Bourret, 1937): MNHNP 1948.153=lectotype, 1938.62, 1948.152=paralectotypes; *T. corticale* (Boulenger, 1903): BM 1947.2.8.39–40=syntypes; *T. horridum* (Boulenger, 1903): BM 1947.2.7.97, 99=syntypes; *T. leporosa* Tschudi, 1838: BM 1947.2.9.19=holotype; *T. phrynoderma* (Ahl, 1927) MSNG 29414=lectotype.